Application Number: 09/932,254
Amendment dated: November 22, 2004
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ELECTED CLAIMS

1. (Original) A method of producing a modified gene fusion construct, the method comprising cojoining two or more heterologous nucleic acid sequences, wherein each heterologous nucleic acid sequence encodes one or more enzymatic domains, and wherein at least one of the two or more heterologous nucleic acid sequences is modified, thereby producing the modified gene fusion construct.
2. (Previously Presented) The method of claim 1, wherein at least one of the two or more heterologous nucleic acid sequences is modified prior to cojoining the two or more heterologous nucleic acid sequences.
3. (Previously Presented) The method of claim 1, wherein at least one of the two or more heterologous nucleic acid sequences is modified after cojoining the two or more heterologous nucleic acid sequences.
4. (Previously Presented) The method of claim 1, wherein the one or more enzymatic domains participate in a same metabolic pathway.
5. (Previously Presented) The method of claim 1, wherein at least one of the two or more heterologous nucleic acid sequence is modified by shuffling at least one nucleic acid sequence.
6. (Original) The method of claim 5, wherein shuffling the at least one nucleic acid sequence comprises recursive sequence recombination.
8. (Previously Presented) The method of claim 1, wherein the two or more heterologous nucleic acid sequences encode enzymatic domains selected from the group consisting of diaminobutyric acid aminotransferase, diaminobutyric acid acetyltransferase, and ectoine synthase.
11. (Previously Presented) The method of claim 1, wherein cojoining the two or more heterologous nucleic acid sequences comprises connecting the two or more heterologous nucleic acid sequences directly to one another.

12. (Previously Presented) The method of claim1, wherein cojoining the two or more heterologous nucleic acid sequences comprises connecting the two or more heterologous nucleic acid sequences with one or more nucleotide linker sequences.

13. (Original) The method of claim 12, wherein the one or more nucleotide linker sequences independently comprise between about three and about 300 nucleotides.

14. (Original) The method of claim 13, wherein the one or more nucleotide linker sequences independently comprise between about 12 to about 90 nucleotides.

15. (Original) The method of claim 12, wherein at least one of the one or more nucleotide linker sequences comprises one or more intron sequences.

16. (Original) The method of claim 12, wherein at least one of the one or more nucleotide linker sequences comprises a nucleotide sequence that encodes a peptide linker.

17. (Original) The method of claim 16, wherein the peptide linker comprises a cleavable peptide sequence or intein sequence.

18. (Original) The method of claim 16, wherein at least about 80% of the amino acid residues in the peptide linker are selected from the group consisting of alanine and glycine residues.

19. (Previously Presented) The method of claim 1, wherein the modified gene fusion construct further comprises one or more transcription regulatory sequences.

20. (Original) The method of claim 19, wherein the one or more transcription regulatory sequences comprises one or more plant transcription regulatory sequences.

32. (Original) A method of producing a gene fusion construct, the method comprising cojoining two or more heterologous nucleic acid sequences that participate in the same metabolic pathway, wherein at least one of the

cojoined nucleic acid sequences is derived from a eukaryote and other cojoined nucleic acid sequence is derived from either a different species of eukaryote or from a prokaryote.

33. (Original) The method of claim 32, wherein at least one of the cojoined nucleic acid sequences is derived from a plant.

34. (Original) The method of claim 32, wherein at least one of the cojoined nucleic acid sequences is derived from a prokaryote.

35. (Original) The method of claim 34, wherein at least one of the cojoined nucleic acid sequences is derived from a plant.

36. (Original) The method of claim 32, wherein at least two of the cojoined nucleic acid sequences are derived from different plant species.

37. (Original) The method of claim 32, wherein the method comprises cojoining three or more heterologous nucleic acid sequences that participate in the same metabolic pathway.

38. (Previously Presented) The method of claim 32, wherein at least one of the cojoined heterologous nucleic acid sequences is modified.

39. (Previously Presented) The method of claim 38, wherein at least one of the cojoined heterologous nucleic acid sequences is shuffled.

41. (Previously Presented) The method of claim 32, wherein the two or more heterologous nucleic acid sequences encode enzymatic domains selected from the group consisting of diaminobutyric acid aminotransferase, diaminobutyric acid acetyltransferase, and ectoine synthase.

44. (Original) The method of claim 32, wherein the cojoined nucleic acid sequences are connected directly to one another.

45. (Original) The method of claim 32, wherein the cojoined nucleic acid sequences are connected to one another with one or more nucleotide linker sequences.

46. (Previously Presented) The method of claim 32, wherein the modified gene fusion construct further comprises one or more transcription regulatory sequences.

47. (Original) The method of claim 46, wherein the one or more transcription regulatory sequences comprises one or more plant transcription regulatory sequences.

80. (Original) A method of producing a gene fusion construct, the method comprising cojoining two or more nucleic acid sequences encoding at least two enzymatic domains, wherein at least one of the nucleic acid is derived from a plant, thereby producing a gene fusion construct.

82. (Previously Presented) The method of claim 80, wherein at least two enzymatic domains are derived from proteins that participate in the same metabolic pathway.

83. (Original) The method of claim 80, wherein at least one of the two or more nucleic acid sequences is modified.

84. (Original) The method of claim 83, wherein at least one of the two or more nucleic acid sequences is modified by shuffling.

85. (Original) The method of claim 84, wherein shuffling the at least one nucleic acid sequence comprises recursive sequence recombination.

86. (Original) The method of claim 80, wherein cojoining the two or more nucleic acid sequences comprises connecting the two or more nucleic acid sequences directly to one another.

87. (Original) The method of claim 80, wherein cojoining the two or more nucleic acid sequences comprises connecting the two or more nucleic acid sequences with one or more nucleotide linker sequences.

88. (Original) The method of claim 87, wherein the one or more nucleotide linker sequences independently comprise between about three and about 300 nucleotides.

89. (Original) The method of claim 88, wherein the one or more nucleotide linker sequences independently comprise between about 12 to about 90 nucleotides.

90. (Original) The method of claim 87, wherein at least one of the one or more nucleotide linker sequences comprises one or more intron sequences.

91. (Original) The method of claim 87, wherein at least one of the one or more nucleotide linker sequences comprises a nucleotide sequence that encodes a peptide linker.

92. (Original) The method of claim 91, wherein the peptide linker comprises a cleavable peptide sequence or an intein sequence.

93. (Previously Presented) The method of claim 91, wherein at least about 80% of amino acid residues in the peptide linker are selected from the group consisting of alanine and glycine residues.

94. (Previously Presented) The method of claim 80, wherein the gene fusion construct further comprises one or more transcription regulatory sequences.

95. (Original) The method of claim 94, wherein the one or more transcription regulatory sequences comprises one or me plant transcription regulatory sequences.

105. (New) The method of claim 80, wherein the at least two enzymatic domains comprise domains from plant enzymes selected from the group consisting of diaminobutyric acid aminotransferase, diaminobutyric acid acetltransferase and ecotoine synthase.